Review

Macromolecular Carrier Systems for Targeted Drug Delivery: Pharmacokinetic Considerations on Biodistribution

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This review article describes the current status and future perspectives of site-specific drug delivery by means of macromolecular carrier systems. Basic aspects and recent advances of targeted delivery of 1) conventional drugs, 2) protein drugs, and 3) gene medicines including antisense oligonucleotides and plasmid DNA, are reviewed from a pharmacokinetic perspective. Successful in vivo application of macromolecular carrier systems requires pharmacokinetic considerations at whole body, organ, cellular and subcellular levels. The integration of simultaneous research progress in the multidisciplinary fields such as biochemistry, cell and molecular biology, pharmacology, and pharmacokinetics will accelerate the emergence of marketed drugs with macromolecular carrier systems.

KEY WORDS: macromolecular carrier; pharmacokinetics; targeting; protein drugs; gene medicines.

INTRODUCTION

For effective therapy with medication, it is necessary to deliver therapeutic agents selectively to their target sites, since most drugs are associated with both beneficial effects and unfavorable actions. This selectivity is best for antitumor drugs because of their extreme cytotoxicity. In general, the lack of selectivity of most conventional drugs is closely related to their pharmacokinetic properties. The in vivo fate of a drug given by a particular administration route is determined by both the physicochemical properties of the drug and anatomical and physiological characteristics of the body. Most conventional drugs diffuse freely throughout the body and show relatively even tissue distribution due to their low molecular weight.

Among the various strategies for site-specific drug delivery, that of macromolecular carriers can be a formidable tool because of their diversity in physicochemical and biological properties and functions (1–3). The rationale for a macromolecular carrier approach in site-specific drug delivery lies in the altered disposition of a carrier-conjugated drug in the body, which is largely dictated by the properties of the carrier and accordingly differs from that of the free drug administered by the same route. In this context, it is important to understand the pharmacokinetics of macromolecules in relation to their physicochemical and biological characteristics.

The progress in biotechnology, such as being able to synthesize DNA constructs containing genes of interest, has effected dramatic changes in therapeutic modalities. While only “xenobiotics” have been used mostly as medicines in conventional drug therapy, the use of endogenous macromolecules and related substances as therapeutic moieties have become increasingly common. The first generation of therapeutic recombinant proteins has already been clinically applied. Efforts have also been made to develop more effective delivery systems for improving the pharmacokinetic properties of proteins drugs and to render proteins more realistic as drug candidates in therapeutics (4). Macromolecular carriers may be useful for some of these protein drugs.

Furthermore, recombinant DNA itself has been used like a “drug” in the novel therapeutic methodology of gene therapy, in which a variety of diseases may be treated by transferring genetic material into specific cells of a patient (5). On the other hand, the inhibition of gene expression using antisense oligonucleotides, which are relatively small synthetic DNA fragments designed to hybridize specific mRNA sequences in target cells, can be considered as a novel type of chemotherapy (6). Macromolecular carriers have been used for the targeted delivery of these DNA drugs “gene medicines”, for therapy at the level of gene expression.

This article reviews the current status and future prospects of targeted drug delivery with macromolecular carrier systems. For the rational design of such targeted drug delivery systems aiming at controlled biodistribution, pharmacokinetic aspects are important (7). In this review, the site-specific delivery of conventional drugs, protein drugs and gene medicines by the use of macromolecular carriers is discussed based on pharmacokinetic considerations at the whole body, organ, and cellular levels.

Design of Drug-Macromolecule Complexes

A variety of natural and synthetic macromolecules have been used as drug carriers. The criteria for choice of macromo-

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Macromolecular Carriers for Drug Targeting

Macromolecular carriers can be summarized as follows. The carrier should (a) be biocompatible, (b) lack intrinsic toxicity and antigenicity, (c) not accumulate in the body, (d) have adequate functional groups for chemical fixation, (e) retain the original specificity for target, and (f) maintain the original activity of the delivered drug until it reaches the site of action. The design of drug-macromolecule complexes can be classified into three categories depending on the drug type.

**Macromolecular Prodrugs**

For most low molecular weight drugs, drug-macromolecular carrier conjugates are classified as macromolecular prodrugs (1). Although some polymer-bound antitumor drugs exhibit activities in the conjugated form without liberating free drugs (8), most conjugates exhibit pharmacological efficacy after conversion to the parent compounds by virtue of an enzyme or chemical lability, or both, before or after reaching the site of action in the body.

In designing macromolecular prodrugs, the pharmacologically active component should (a) show enough activity at relatively low doses to decrease the load of a carrier macromolecule (cytotoxic agents are often used for this reason), (b) be chemically stable in the conjugated form until released, and (c) have adequate functional groups in its molecular structure for chemical fixation (1). In addition, the chemical and/or biological stability of the linkage between the drug and the macromolecular carrier should be considered since their pharmacological effectiveness requires the release of the free drug from the conjugate. Generally, drugs are coupled directly to functional groups in a macromolecular carrier or to spacers introduced to a macromolecular backbone via covalent bonds, using various cross-linking agents. Functional groups in the drug and the carrier molecule used for chemical coupling include amino, carboxyl, hydroxyl groups, and free thiol groups. Most of the reactions are performed in aqueous media under mild conditions to avoid denaturation of the parent drug and the macromolecular carrier. These linkages between the drug and the carrier must be designed to be cleavable at an appropriate rate to act as prodrugs. When the linkage is to be cleaved by enzymatic reaction(s), animal species differences in the type and activity of the enzyme(s) should be considered.

**Protein-macromolecule Conjugates**

Macromolecular carriers have been used to chemically modify protein drugs to improve their pharmaceutical, pharmacokinetic and immunological properties. In general, protein drugs are chemically conjugated to macromolecular modifiers and the conjugates, hybrids of two macromolecules, can have unique biological activities unlike those of macromolecular prodrugs. In this case, proteins must have appropriate functional groups, solubility and stability for chemical modification and conjugation. The molecular weight of modifiers and extent of conjugation should be selected to maintain the biological activities of the protein.

Occasionally, conjugation with macromolecular carriers attenuates the biological activities of protein drugs by means of an intrinsic conformational change and/or restricted accessibility to target molecules due to steric hindrance. Enzymes have been most widely used since their substrates, which are usually highly diffusible small molecules, should undergo relatively effective reactions catalyzed by the enzyme even after modification (9). Although steric hindrance should be considered especially for protein drugs that require receptor binding to exert their therapeutic action, several conjugated cytokines can maintain their activities (10).

In addition to macromolecular conjugation, direct chemical modification with small functional moieties that alter the physiological and biological properties of proteins would be useful. These moieties include positively or negatively charged groups, lipophilic groups and sugars. The basic concept similar to macromolecular conjugation can be applied to the design of protein drugs that will be directly modified with these small molecules.

**Gene Medicines Complexed with Macromolecular Carriers**

For successful therapy with gene medicines involving recombinant plasmid DNA and antisense oligonucleotides, it is necessary to develop carrier systems that deliver these materials to the target intracellular site. In general, gene medicines have substantial problems as polyanionic DNA molecules. These include susceptibility to degradation by nucleases and low membrane permeability. The introduction of an appropriate macromolecular counterpart as a carrier of gene medicines would be one useful way to circumvent these problems.

Macromolecular carriers have been covalently attached to oligonucleotides (11). However, the use of non-covalent electrostatic interaction between DNA and polycations is more common (12). Current bifunctional macromolecular conjugates consist of two components, a target recognizable macromolecule and a polycation. The former portion would direct the conjugate to its target cells and the polycationic region can bind DNA based on electrostatic interaction and neutralize anionic charges. Small target recognition elements such as sugars have been introduced directly to polycations. Various ligands that can bind to receptors on the specific cell types such as glycopolymers, transferrin, and insulin, and monoclonal antibodies capable of recognizing cellular epitopes have been used as the targeting component. On the other hand, poly-L-lysine is the most common polycation for the hybrid carrier.

Many factors should be considered in designing a carrier system. These include (a) the choice of targeting moiety, (b) the molecular weight of poly-L-lysine (or other polycation), (c) the coupling ratio of these components, (d) mixing molar ratio of the carrier and DNA, (e) conditions such as ionic strength for mixing of the carrier and DNA (for a review, see ref. 12). Apparently, these factors are dependent on the type and size of the DNA.

**Targeting and Macromolecular Carriers**

Site-specific drug delivery is broadly categorized as passive and active targeting (13). "Passive targeting" refers to the exploitation of the natural (passive) disposition profiles of a drug carrier, which is passively determined by its physicochemical properties relative to the anatomical and physiological characteristics of the body as will be discussed later. "Active targeting" refers to the alterations of the natural disposition of a drug or carrier, directing it to specific cells, tissues, or organs. Ligands or monoclonal antibodies which can bind specifically to the surface of target cells, are used for this purpose. Active targeting seems to be much more attractive than passive tar-